

Untargeted metabolomics to reveal red versus white meat-associated gut metabolites in a prudent and Western dietary context

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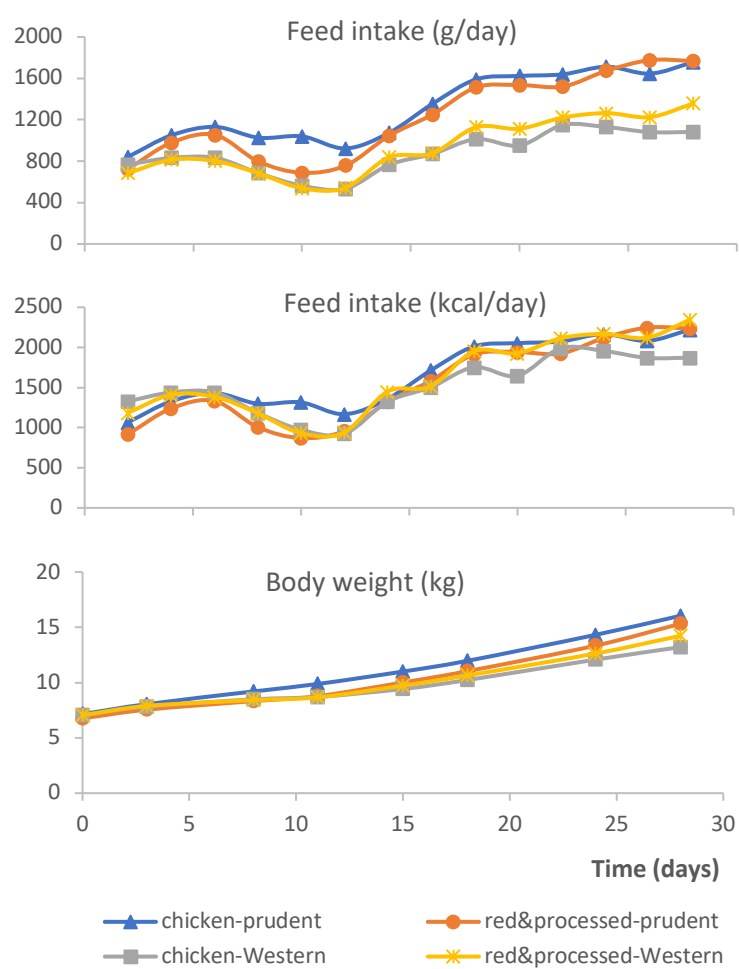


Figure S1. Feed intake in g/day, kcal/day and body weight during the experimental feeding period (n = 8 per diet)

Table S1. Weight and dry matter (mean \pm standard deviation) of the luminal content of the small intestine and colon of the pigs subjected to four dietary treatments (n = 8 per dietary treatment, except n = 6 for colon content of pigs fed the chicken-Western diet).

	prudent		Western	
	chicken	red&processed	chicken	red&processed
luminal content small intestine				
weight (g)	220 \pm 87	310 \pm 158	225 \pm 66	222 \pm 73
dry matter (%)	10.3 \pm 1.7	9.0 \pm 1.4	8.0 \pm 1.8	9.4 \pm 1.6
luminal content colon				
weight (g)	115 \pm 52	108 \pm 55	83 \pm 39	85 \pm 45
dry matter (%)	16.8 \pm 5.2	17.4 \pm 4.3	17.3 \pm 3.9	15.7 \pm 2.6

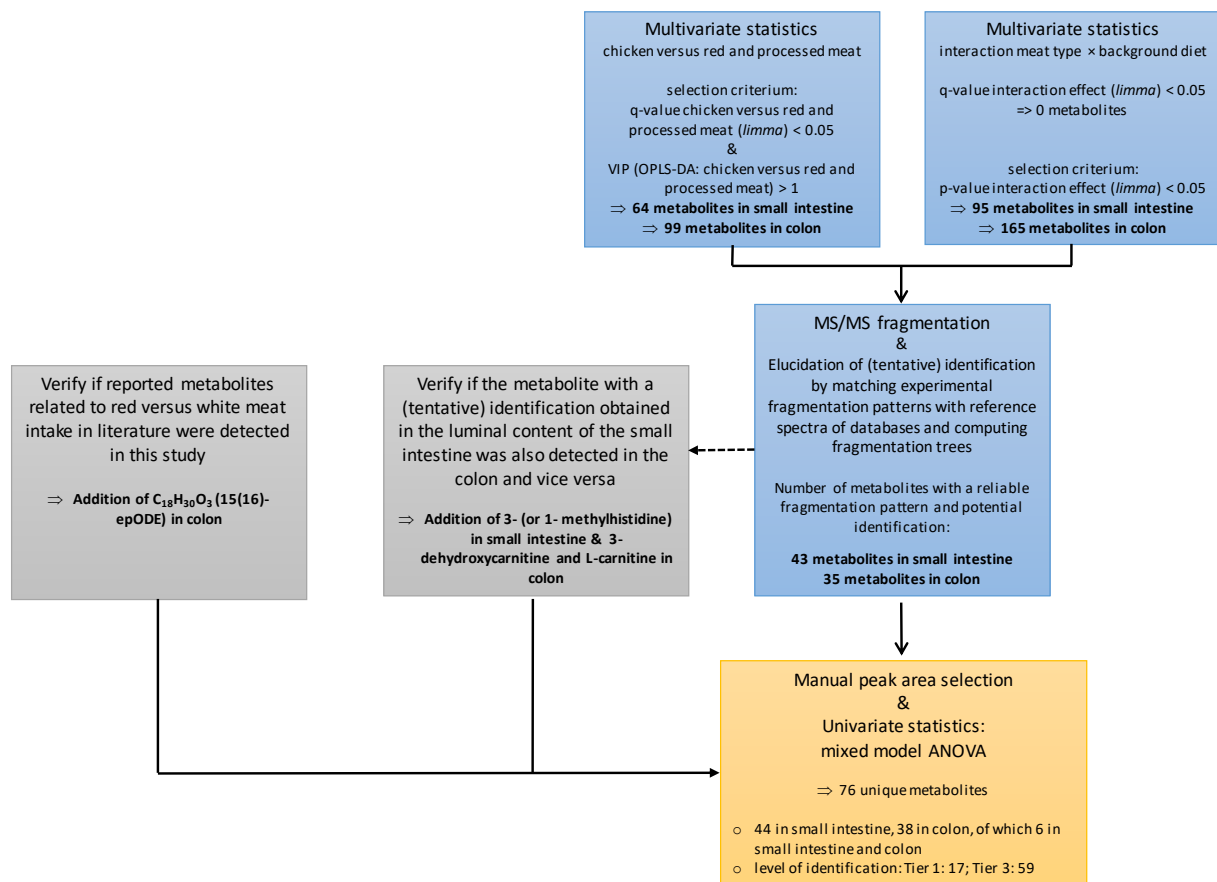


Figure S2. Workflow illustrating the selection of metabolites.

Commercial standards

The authentic standards of cadaverine, L-carnitine, acetyl-L-carnitine (C2-carnitine), propionyl-L-carnitine (C3-carnitine), butyryl-L-carnitine and isobutyryl-L-carnitine (C4-carnitine), valeryl-L-carnitine and isovaleryl-L-carnitine (C5-carnitine), hexanoyl-L-carnitine (C6-carnitine), decanoyl-L-carnitine (C10-carnitine), tetradecanoyl-L-carnitine (C14-carnitine), hexadecanoyl-L-carnitine (C16-carnitine), trans-2-hexadecenoyl-L-carnitine (C16:1-carnitine), octadecanoyl-L-carnitine (C18-carnitine), octadecenoyl-L-carnitine (C18:1-carnitine), undecanedioic acid, 1-oleoylglycerol (oleoylglycerol), 1-stearoylglycerol (stearoylglycerol), 9(S)-hydroxy-10E,12Z-octadecadienoic acid (HODE) and D-pipecolic acid were purchased from Sigma-Aldrich (Overijse, Belgium), whereas 1-palmitoylglycerol (palmitoylglycerol) and 13(S)-Hydroperoxy-9Z,11E-octadecadienoic acid (HPODE) were obtained from Santa Cruz Biotechnology Inc. (Heidelberg, Germany). Anserine and carnosine were gifts from a collaborating laboratory.

Standards were obtained for 22 metabolites and the identity of 17 metabolites was confirmed. The identity of the remaining five (trans-2-hexadecenoyl-L-carnitine (C16:1-carnitine), 1-palmitoylglycerol (palmitoylglycerol), 1-stearoylglycerol (stearoylglycerol), 13(S)-hydroperoxy-9Z,11E-octadecadienoic acid (HPODE) and 9(S)-hydroxy-10E,12Z-octadecadienoic acid (HODE)) was not confirmed due to differences in retention time (> 0.2 min) and/or different MS/MS spectra. However, they were retained in the list as putatively characterized metabolites based on good CSI:FingerID and Metfrag scores, previously annotation of certain metabolites in relation with meat in literature, and/or the putative identification of multiple compounds of the same classes. The retention time of the standard trans-2-hexadecenoyl-L-carnitine did not correspond with the retention time of the compound that was tentatively identified as C16:1-carnitine. However, the presence of the specific 85.029 Da fragment in its fragmentation profile increases the likelihood that this unidentified metabolite is an acylcarnitine (and an isomer of trans-2-hexadecenoyl-L-carnitine). For palmitoylglycerol and stearoylglycerol, the fragmentation patterns of the standards and the unidentified metabolites corresponded to a great extent, but not the retention times. For another group of compounds, the linoleic acid derivatives, (of which the standards 13(S)-hydroperoxy-9Z,11E-octadecadienoic acid and 9(S)-hydroxy-10E,12Z-octadecadienoic acid were available) multiple potential identity candidates are present and difficult to identify, as multiple possible structures with an identical molecular mass, molecular formula and very similar fragmentation patterns for this class of metabolites exist.

Fragmentation patterns of butyryl-L-carnitine (retention time: 6.27 min) and isobutyryl-L-carnitine (retention time: 6.08 min), and of valeryl-L-carnitine (retention time: 8.20 min) and isovaleryl-L-carnitine (retention time: 8.02 min) were very similar. However, based on the retention times of the standards and the metabolites in the samples, eluting at 6.28 min and 8.03 min, the metabolites C4-carnitine and C5-carnitine could be respectively identified as butyryl-L-carnitine and isovaleryl-L-carnitine.

Table S2. Retention time and fragmentation pattern of the commercially available standards

(Tentatively) characterized metabolites	Corresponding standards	RT sample (min)	RT standard (min)	MS/MS fragment ions standards (relative abundance)
Standards compared with metabolites detected in small intestinal digests				
Cadaverine ^a	cadaverine	0.73	0.68	102.128 (100) 74.097 (16) 103.131 (13) 58.066 (6) 72.081 (2)
Anserine ^a	anserine	0.80	0.80	102.128 (100) 142.948 (40) 241.129 (25) 170.092 (15) 109.076 (13)
L-carnitine ^a	L-carnitine	0.85	0.84	162.112 (100) 103.039 (17) 60.082 (16) 102.092 (4) 85.029 (3)
C2-carnitine ^a	acetyl-L-carnitine	1.27	1.26	85.029 (100) 204.123 (39) 60.082 (17) 145.050 (14) 144.102 (3)
C3-carnitine ^a	propionyl-L-carnitine	3.06	3.03	85.029 (100) 218.138 (28) 159.065 (16) 60.082 (13) 86.032 (2)
C4-carnitine ^a	butyryl-L-carnitine	6.28	6.27	85.029 (100) 232.154 (24) 173.081 (16) 60.082 (12) 144.101 (2)
C5-carnitine ^a	isovaleryl-L-carnitine	8.01	8.02	85.029 (100) 246.169 (26) 187.096 (19) 60.082 (12) 85.065 (4)
C6-carnitine ^a	hexanoyl-L-carnitine	9.82	9.80	85.029 (100) 260.185 (21) 201.112 (14) 60.082 (11) 99.081 (4)
Carnosine ^a	carnosine	0.80	0.81	210.087 (100) 156.077 (97) 110.072 (83) 227.114 (64) 181.108 (33)
C10-carnitine ^a	decanoyl-L-carnitine	11.10	11.10	85.029 (100) 316.248 (19) 60.082 (12) 257.174 (8) 155.143 (3)
C14-carnitine ^a	tetradecanoyl-L-carnitine	12.01	11.99	85.029 (100) 372.310 (16) 60.082 (12) 313.237 (6) 144.102 (3)
C16:1-carnitine ^b	trans-2-hexadecenoyl-L-carnitine	12.17	12.43	85.029 (100) 398.326 (11) 339.252 (9) 237.221 (9) 255.232 (9)
C16-carnitine ^a	hexadecanoyl-L-carnitine	12.83	12.81	85.029 (100) 400.342 (20) 60.082 (13) 341.269 (5) 144.102 (3)
C18:1-carnitine ^a	octadecenoyl-L-carnitine	12.96	12.97	85.029 (100) 426.358 (17) 60.082 (14) 144.102 (3) 95.086 (2)
C18-carnitine ^a	octadecanoyl-L-carnitine	13.10	13.11	85.029 (100) 428.372 (16) 60.082 (13) 369.299 (3) 144.102 (2)
Palmitoylglycerol ^b	1-palmitoylglycerol	15.28	15.48	313.273 (100) 57.071 (44) 95.086 (38) 293.237 (31) 71.086 (31)
Oleoylelycerol ^a	1-oleoylelycerol	14.82	14.82	95.086 (100) 69.071 (95) 81.071 (83) 83.086 (72) 121.102 (63)
Stearoylelycerol ^b	1-stearoylelycerol	14.13	13.65	341.304 (100) 95.086 (36) 57.071 (34) 97.102 (30) 109.101 (27)
Standards compared with metabolites detected in colon digests				
Undecanedioic acid ^a	undecanedioic acid	11.05	11.04	135.117 (100) 153.127 (63) 69.071 (48) 97.065 (35) 217.143 (27)
C ₁₈ H ₃₂ O ₃ (HODE) ^b	9(S)-hydroxy-10E,12Z-octadecadienoic acid	13.88	13.38	277.217 (100) 295.228 (99) 171.102 (64) 296.232 (9) 278.221 (8)
C ₁₈ H ₃₂ O ₄ (HPODE) ^b	13(S)-Hydroperoxy-9Z,11E-octadecadienoic acid	12.57	13.41	113.096 (100) 293.212 (47) 195.138 (17) 57.033 (15) 59.012 (13)
D-pipecolic acid ^a	D-pipecolic acid	1.26	1.22	84.081 (100) 130.086 (34) 85.084 (3) 131.090 (1) 84.045 (1)

a) Identification was confirmed with commercial standards. b) The identity of the detected metabolite does not correspond with the available specific standard, but is likely an isomer of the available standard, which is covered by the nomenclature used in the list.

Note that retention time of the metabolites in the samples shown in this table may deviate from the retention time in Tables S2 and S3, due to the analysis of the samples with the UHPLC-Orbitrap-HRMS procedure (Vanden Bussche et al., Anal. Chem. 2015, 87, 10927-10934) and the additional fragmentation analysis with the slightly adapted UHPLC-Q-Orbitrap-HRMS procedure (De Paepe et al., Anal. Chim. Acta 2018, 1033, 108-118).

Table S3. Retention time and monoisotopic mass of metabolites (n = 64) that were influenced by meat intake in the luminal content of the small intestine and obtained by multivariate statistics. Benjamini Hochberg adjusted p-values (q-values) were obtained from Empirical Bayes moderated t-statistics in *limma* investigating the main effect meat.

monoisotopic mass	RT (min)	higher after intake of ...	VIP	q-value	monoisotopic mass	RT (min)	higher after intake of ...	VIP	q-value
261.1206	1.6	red&processed	6.9	1.72E-17	312.2657	13.6	red&processed	2.5	1.25E-02
161.1048	0.9	red&processed	7.1	1.72E-17	341.2556	9.8	red&processed	2.8	1.25E-02
203.1153	1.4	red&processed	6.3	2.64E-16	483.3006	10.0	chicken	3.4	1.38E-02
145.1100	1.0	red&processed	5.3	4.00E-12	85.0892	0.8	chicken	2.9	1.38E-02
231.1466	5.2	red&processed	5.8	1.06E-11	113.0588	1.0	chicken	2.8	1.38E-02
247.1414	1.7	red&processed	5.5	1.61E-11	367.2714	9.9	red&processed	2.6	1.38E-02
400.1950	6.6	chicken	4.6	2.09E-10	203.0976	8.5	red&processed	3.1	1.38E-02
217.1310	2.7	red&processed	5.2	3.18E-10	256.1054	3.0	red&processed	2.1	1.39E-02
108.5713	0.8	chicken	4.9	4.08E-10	140.0582	1.0	chicken	3.0	1.41E-02
259.1778	8.2	red&processed	4.8	4.08E-10	346.1481	2.6	chicken	1.8	1.51E-02
120.0609	0.9	chicken	5.1	5.34E-10	102.1156	0.8	chicken	2.9	1.59E-02
240.1216	0.9	chicken	5.5	1.13E-09	201.1005	14.6	red&processed	2.0	1.59E-02
214.1313	1.8	chicken	4.1	1.26E-09	282.1321	1.4	chicken	2.4	1.82E-02
315.2401	9.5	red&processed	5.0	1.26E-09	399.3339	11.1	red&processed	2.7	1.99E-02
244.1418	1.6	red&processed	4.0	1.42E-09	509.3837	13.0	red&processed	2.5	2.20E-02
312.1426	1.4	chicken	3.7	2.87E-09	352.2605	12.5	red&processed	2.3	2.26E-02
114.0428	1.8	red&processed	5.9	1.43E-08	519.2168	5.2	chicken	1.9	2.43E-02
129.0899	1.4	chicken	4.7	7.83E-08	479.3364	11.9	red&processed	2.8	2.43E-02
172.1321	1.7	chicken	4.9	1.98E-07	292.2030	11.3	red&processed	3.0	2.45E-02
245.1622	6.6	red&processed	4.8	3.19E-07	361.1477	5.4	chicken	2.0	2.76E-02
447.1957	3.3	chicken	3.5	4.68E-06	483.3006	9.5	chicken	2.9	3.27E-02
303.1423	5.5	chicken	2.9	3.51E-05	579.8003	7.0	red&processed	2.6	4.40E-02
313.2247	9.4	red&processed	2.8	6.39E-05					
264.0775	2.7	chicken	2.8	2.40E-04					
507.3680	12.9	red&processed	3.4	2.63E-04					
369.2870	10.2	red&processed	3.9	2.63E-04					
371.3026	10.5	red&processed	3.3	6.05E-04					
343.2713	10.0	red&processed	3.4	7.04E-04					
427.3651	11.6	red&processed	3.6	7.04E-04					
397.3182	10.7	red&processed	3.7	1.33E-03					
451.3056	12.3	red&processed	2.9	2.22E-03					
226.1061	0.9	red&processed	3.2	2.42E-03					
114.0621	0.9	chicken	2.5	2.48E-03					
113.0532	0.8	red&processed	3.2	2.50E-03					
463.3285	10.3	chicken	2.8	4.01E-03					
425.3494	11.2	red&processed	3.3	4.42E-03					
255.6132	5.4	chicken	2.4	5.35E-03					
280.1053	5.8	chicken	2.4	6.06E-03					
330.2762	13.6	red&processed	2.9	6.29E-03					
423.3340	10.9	red&processed	2.9	9.35E-03					
294.2187	10.9	red&processed	3.1	1.22E-02					
465.2900	10.0	chicken	3.2	1.23E-02					

Table S4. Retention time and monoisotopic mass of metabolites (n = 99) that were influenced by meat intake in the luminal content of the colon and obtained by multivariate statistics. Benjamini Hochberg adjusted p-values (q-values) were obtained from Empirical Bayes moderated t-statistics in *limma* investigating the main effect meat.

monoisotopic mass	RT (min)	higher after intake of ...	VIP	q-value	monoisotopic mass	RT (min)	higher after intake of ...	VIP	q-value
312.1431	1.4	chicken	5.7	9.91E-13	608.3211	9.4	chicken	2.9	2.18E-03
244.1421	1.5	red&processed	5.9	1.21E-12	227.1158	4.0	chicken	2.5	2.31E-03
213.1113	2.0	chicken	6.0	4.99E-12	226.1204	9.1	chicken	2.5	3.01E-03
141.0618	1.0	chicken	6.0	1.41E-11	216.1361	9.5	chicken	2.9	3.05E-03
140.0584	1.0	chicken	5.7	7.46E-11	242.1881	11.9	red&processed	2.9	3.28E-03
161.0686	1.9	chicken	5.3	7.56E-11	201.1364	3.2	chicken	2.2	3.57E-03
140.0585	1.3	chicken	5.6	2.22E-10	296.1735	6.9	red&processed	3.1	4.01E-03
237.1000	3.8	chicken	5.0	2.22E-10	155.0693	0.9	chicken	2.2	4.09E-03
126.0793	1.0	chicken	5.4	3.19E-10	239.1268	2.5	chicken	2.8	4.69E-03
212.1273	3.9	chicken	4.7	2.54E-08	128.0586	1.2	chicken	2.0	5.56E-03
170.0690	1.0	chicken	4.8	3.92E-08	147.0894	1.0	red&processed	2.5	6.04E-03
202.0952	2.3	chicken	3.9	1.59E-07	305.1682	9.5	chicken	2.1	6.20E-03
168.0772	1.0	chicken	3.7	2.17E-07	188.1043	9.0	chicken	2.5	7.35E-03
402.2265	9.1	chicken	4.6	4.80E-07	230.1378	1.8	chicken	2.9	9.59E-03
141.0902	2.1	chicken	3.5	5.65E-07	96.0689	1.7	chicken	1.8	1.11E-02
231.1834	7.9	chicken	5.2	1.46E-06	160.1211	1.0	chicken	1.8	1.13E-02
256.1169	1.5	chicken	3.1	5.98E-06	329.2563	9.0	red&processed	2.3	1.13E-02
312.1430	1.0	chicken	2.9	1.33E-05	292.0555	1.0	chicken	1.6	1.18E-02
159.1006	1.7	chicken	4.2	1.84E-05	197.0800	1.0	chicken	2.1	1.31E-02
159.1008	1.9	chicken	4.5	2.49E-05	169.0851	0.9	chicken	2.0	1.40E-02
246.1466	9.0	chicken	3.1	3.48E-05	145.1102	1.4	chicken	2.2	1.55E-02
278.2245	10.2	chicken	3.3	3.48E-05	352.2223	9.6	red&processed	2.5	1.55E-02
360.1974	10.1	chicken	4.6	3.48E-05	184.0960	1.4	chicken	1.9	1.58E-02
431.2339	9.4	chicken	3.2	5.28E-05	156.0535	2.4	chicken	2.0	1.62E-02
126.0667	1.2	chicken	3.0	5.79E-05	169.0850	2.0	chicken	1.9	1.83E-02
432.2375	9.4	chicken	2.8	6.48E-05	201.1001	4.9	chicken	1.6	1.83E-02
211.0956	1.0	chicken	3.1	6.68E-05	109.0528	1.0	chicken	1.6	1.91E-02
281.1296	2.9	chicken	2.9	7.08E-05	221.1262	1.0	chicken	2.2	2.11E-02
313.1999	6.0	chicken	3.8	7.32E-05	312.2301	11.0	red&processed	2.6	2.13E-02
244.1311	9.1	chicken	3.3	1.24E-04	611.3404	9.5	chicken	1.9	2.24E-02
304.1605	9.4	chicken	2.7	2.00E-04	83.0374	0.9	chicken	1.7	2.36E-02
157.1102	3.9	chicken	2.8	2.66E-04	329.2564	10.9	red&processed	2.2	2.44E-02
160.0848	3.1	chicken	3.7	2.80E-04	181.0741	9.4	chicken	1.6	2.53E-02
213.1113	3.9	chicken	2.9	3.05E-04	347.2670	9.7	red&processed	2.0	2.80E-02
171.0895	5.4	chicken	4.0	3.17E-04	185.0800	1.0	chicken	2.1	2.82E-02
342.2041	9.7	chicken	2.5	3.78E-04	289.0617	4.4	chicken	2.2	2.94E-02
228.1360	9.0	chicken	2.5	6.54E-04	271.1611	6.2	chicken	1.8	2.98E-02
96.0689	1.2	chicken	2.4	7.05E-04	208.1098	9.1	chicken	1.9	3.18E-02
157.1101	1.4	red&processed	2.7	7.15E-04	129.0790	1.0	red&processed	2.7	3.24E-02
207.0895	3.3	chicken	3.0	8.60E-04	210.1255	9.6	chicken	1.7	3.24E-02
179.0946	6.9	red&processed	3.4	1.25E-03	156.0787	8.0	chicken	2.1	3.28E-02
180.1149	10.1	red&processed	2.1	1.25E-03	267.0200	7.3	chicken	1.8	3.42E-02
170.0942	9.0	chicken	2.9	1.38E-03	429.2184	9.2	chicken	1.8	3.43E-02
245.1010	1.4	red&processed	2.8	1.39E-03	610.3367	9.5	chicken	2.4	3.43E-02
239.1157	5.1	chicken	2.2	1.40E-03	217.1313	3.3	red&processed	2.4	3.59E-02
147.0683	9.1	red&processed	2.7	1.54E-03	110.0369	1.2	chicken	1.5	3.76E-02
202.1064	1.0	chicken	2.2	1.86E-03	85.0530	0.9	chicken	2.0	3.88E-02
207.0895	2.4	chicken	2.8	2.02E-03	330.2408	9.7	red&processed	1.9	3.94E-02
223.0299	7.3	chicken	2.6	2.13E-03	413.2235	9.4	chicken	1.9	3.97E-02
					606.3062	9.3	chicken	1.6	4.19E-02

Table S5. Retention time and monoisotopic mass of metabolites (n = 95) in the luminal content of the small intestine that were affected by the combination of both the meat type and background diet. Non-adjusted p-values were obtained from Empirical Bayes moderated t-statistics in *limma* investigating the interaction effect meat×background diet.

monoisotopic mass	RT (min)	p-value	monoisotopic mass	RT (min)	p-value	monoisotopic mass	RT (min)	p-value
579.8003	7.0	1.86E-03	438.2973	11.1	2.04E-02	336.2656	13.1	3.56E-02
502.7633	6.8	2.29E-03	159.0891	6.0	2.05E-02	460.2788	11.1	3.63E-02
100.0160	1.9	5.29E-03	151.0491	3.0	2.13E-02	126.0427	4.5	3.67E-02
118.0254	1.9	5.38E-03	267.0961	3.0	2.14E-02	388.2968	11.6	3.68E-02
486.1702	1.2	5.81E-03	356.2918	13.8	2.18E-02	174.1112	0.9	3.70E-02
118.0265	1.9	6.19E-03	373.0550	10.6	2.19E-02	192.0624	1.0	3.72E-02
74.0356	1.9	6.33E-03	241.1057	1.8	2.26E-02	126.0540	0.6	3.76E-02
148.0361	1.8	7.44E-03	122.0672	14.8	2.28E-02	200.1520	10.0	3.81E-02
370.2863	12.7	7.47E-03	192.0211	1.5	2.30E-02	212.9721	1.9	3.82E-02
519.5007	15.3	7.59E-03	785.0856	10.3	2.34E-02	85.0659	15.3	3.88E-02
313.2247	9.4	8.15E-03	93.9341	0.8	2.44E-02	102.0429	1.2	4.05E-02
208.0486	15.0	9.57E-03	246.1211	4.7	2.59E-02	401.2768	9.4	4.06E-02
262.0798	1.4	1.07E-02	334.0644	1.8	2.69E-02	174.1001	2.0	4.08E-02
156.0426	14.1	1.10E-02	156.0523	9.6	2.69E-02	407.3028	10.9	4.08E-02
328.2395	9.3	1.13E-02	358.3075	14.7	2.74E-02	494.3706	9.7	4.13E-02
445.3185	9.7	1.13E-02	392.2920	11.2	2.77E-02	406.2208	5.1	4.14E-02
344.3283	13.0	1.18E-02	281.9898	9.6	2.81E-02	255.6132	5.4	4.15E-02
170.1416	9.3	1.21E-02	356.2704	9.5	2.89E-02	357.2736	9.6	4.16E-02
393.2955	11.1	1.43E-02	374.2809	10.3	2.95E-02	162.0889	6.5	4.23E-02
136.0382	3.2	1.50E-02	175.0952	0.9	3.01E-02	594.3407	9.1	4.29E-02
88.0150	9.1	1.59E-02	485.2741	9.5	3.04E-02	73.0893	9.0	4.32E-02
370.2862	11.6	1.61E-02	129.5257	11.5	3.05E-02	130.0456	1.6	4.33E-02
352.2605	12.5	1.65E-02	464.5788	7.3	3.06E-02	464.2957	9.5	4.37E-02
137.0474	6.2	1.69E-02	119.0162	1.7	3.07E-02	201.0214	1.5	4.39E-02
106.0124	1.4	1.78E-02	314.2449	10.5	3.09E-02	385.2455	9.6	4.55E-02
328.2607	12.8	1.82E-02	231.6211	5.3	3.12E-02	246.1210	2.0	4.56E-02
65.0256	15.3	1.88E-02	82.0520	9.6	3.16E-02	166.0620	8.1	4.61E-02
244.0690	1.8	1.89E-02	217.1310	2.7	3.18E-02	86.0481	1.7	4.63E-02
230.1626	6.0	1.96E-02	132.0775	7.3	3.19E-02	85.0892	0.8	4.81E-02
555.3029	9.5	1.98E-02	137.0837	2.1	3.32E-02	480.3187	9.5	4.82E-02
252.0853	3.2	2.02E-02	242.0897	4.5	3.37E-02	388.2970	12.7	4.92E-02
			158.0688	0.9	3.45E-02	463.2923	9.5	5.00E-02

Table S6. Retention time and monoisotopic mass of metabolites (n = 165) in the luminal content of the colon that were affected by the combination of both the meat type and background diet. Non-adjusted p-values were obtained from Empirical Bayes moderated t-statistics in *limma* investigating the interaction effect meat×background diet.

monoisotopic mass	RT (min)	p-value	monoisotopic mass	RT (min)	p-value	monoisotopic mass	RT (min)	p-value
298.2505	10.6	7.11E-05	160.1293	1.5	1.27E-02	270.1578	6.2	3.14E-02
99.0685	4.2	1.20E-04	354.2554	10.0	1.29E-02	464.1869	6.7	3.15E-02
287.1843	6.9	4.09E-04	308.1986	9.4	1.29E-02	153.0426	5.0	3.20E-02
316.2611	10.6	4.26E-04	345.2514	9.7	1.34E-02	130.1105	15.7	3.33E-02
317.1586	4.9	4.39E-04	332.1960	10.5	1.42E-02	167.1057	1.6	3.33E-02
264.1472	4.2	4.94E-04	226.1680	9.2	1.43E-02	84.0326	9.8	3.34E-02
121.0504	4.3	6.32E-04	210.1255	9.6	1.46E-02	130.0823	1.4	3.37E-02
314.2457	10.9	9.68E-04	355.2588	10.0	1.50E-02	324.1935	9.7	3.37E-02
336.2274	10.9	1.03E-03	262.1316	6.8	1.52E-02	215.1521	9.6	3.38E-02
100.0719	4.3	1.21E-03	83.0374	0.9	1.52E-02	110.0320	10.5	3.42E-02
260.1523	9.2	1.87E-03	131.0210	4.4	1.53E-02	97.0065	1.9	3.45E-02
118.0630	3.5	1.96E-03	228.1360	9.0	1.57E-02	312.2298	11.2	3.45E-02
246.1466	9.0	2.03E-03	338.2430	10.6	1.58E-02	153.0900	1.4	3.52E-02
183.1007	4.5	2.25E-03	67.0048	7.8	1.63E-02	205.0735	8.5	3.54E-02
189.1364	2.4	2.32E-03	326.2092	9.4	1.71E-02	194.0283	2.0	3.57E-02
216.1109	1.8	2.60E-03	173.0687	1.5	1.73E-02	406.2715	10.0	3.59E-02
296.2349	10.9	2.63E-03	187.1207	1.9	1.79E-02	404.2558	10.0	3.59E-02
328.0948	8.7	3.10E-03	245.1010	1.4	1.88E-02	156.0787	8.0	3.62E-02
445.3188	8.9	3.14E-03	407.2748	10.0	1.90E-02	154.0741	1.0	3.66E-02
344.2201	9.3	3.19E-03	408.2870	10.0	2.06E-02	289.1272	1.0	3.68E-02
105.0322	13.7	3.20E-03	281.1296	2.9	2.09E-02	144.1261	1.4	3.70E-02
276.1473	7.4	3.55E-03	201.1001	4.9	2.11E-02	230.0900	1.5	3.71E-02
189.1364	2.9	4.24E-03	455.2964	10.0	2.12E-02	217.1313	3.3	3.76E-02
127.0746	13.5	4.50E-03	314.1701	3.0	2.13E-02	348.1909	9.9	3.83E-02
343.1740	5.5	4.96E-03	132.0979	1.0	2.15E-02	483.3926	15.1	3.83E-02
344.2349	10.0	5.44E-03	115.0634	1.8	2.19E-02	159.1258	1.0	3.89E-02
276.1472	7.8	5.47E-03	289.0617	4.4	2.20E-02	390.2764	10.0	3.91E-02
170.0942	9.0	5.48E-03	373.2694	10.0	2.21E-02	454.2928	10.0	3.94E-02
212.1524	8.4	5.81E-03	170.1055	5.7	2.21E-02	259.1166	1.6	4.07E-02
244.1311	9.1	6.01E-03	139.0996	8.8	2.23E-02	235.0957	5.5	4.11E-02
278.2244	10.9	6.02E-03	305.1682	9.5	2.34E-02	293.6432	10.0	4.11E-02
246.1366	8.7	6.15E-03	182.0432	1.5	2.35E-02	160.1000	6.2	4.21E-02
212.1524	7.9	6.45E-03	322.1524	8.2	2.36E-02	112.0274	0.7	4.24E-02
268.0806	3.8	7.33E-03	184.1212	6.8	2.37E-02	89.0103	0.7	4.25E-02
100.0526	3.5	7.60E-03	372.2659	10.0	2.40E-02	509.3633	13.7	4.26E-02
290.1881	9.4	7.66E-03	161.1051	1.4	2.55E-02	242.1265	6.7	4.27E-02
142.0630	6.7	8.52E-03	439.3659	15.2	2.61E-02	207.1258	5.1	4.28E-02
160.0730	6.7	8.56E-03	92.0476	1.2	2.62E-02	131.0695	1.0	4.29E-02
329.1584	4.6	8.81E-03	174.0887	8.1	2.64E-02	326.0007	15.0	4.32E-02
227.1158	4.0	8.93E-03	228.1361	8.1	2.66E-02	172.0847	1.0	4.34E-02
124.0525	6.7	9.40E-03	169.0851	0.9	2.71E-02	256.1672	10.1	4.35E-02
188.1043	9.0	9.47E-03	389.2642	10.0	2.72E-02	231.1834	7.9	4.36E-02
431.3029	10.0	9.63E-03	90.9976	0.7	2.75E-02	262.0986	4.0	4.39E-02
157.1102	3.9	9.94E-03	195.0895	2.6	2.77E-02	446.3006	14.5	4.47E-02
272.1294	10.0	9.99E-03	143.0735	6.2	2.81E-02	327.2407	10.6	4.50E-02
100.0526	8.7	1.01E-02	329.2563	9.0	2.82E-02	315.9127	0.9	4.50E-02
285.1690	6.6	1.03E-02	340.1631	6.4	2.88E-02	244.1788	6.9	4.56E-02
423.2978	10.2	1.09E-02	158.1418	2.2	2.95E-02	136.0636	1.0	4.60E-02
89.0842	15.6	1.10E-02	114.0318	3.3	2.96E-02	175.1207	1.7	4.66E-02
186.1004	1.6	1.15E-02	289.1272	1.4	3.03E-02	60.0004	15.4	4.69E-02
444.2641	10.0	1.17E-02	292.1786	10.0	3.05E-02	221.1262	1.0	4.73E-02
230.1378	1.8	1.19E-02	70.0158	8.3	3.07E-02	244.1788	6.6	4.75E-02
200.1526	10.1	1.20E-02	254.2245	11.7	3.08E-02	157.0739	5.0	4.76E-02

175.0632	9.0	4.80E-02	237.1000	3.8	4.88E-02
209.1051	1.8	4.81E-02	244.1422	4.8	4.89E-02
70.0157	11.6	4.83E-02	126.1158	15.6	4.98E-02

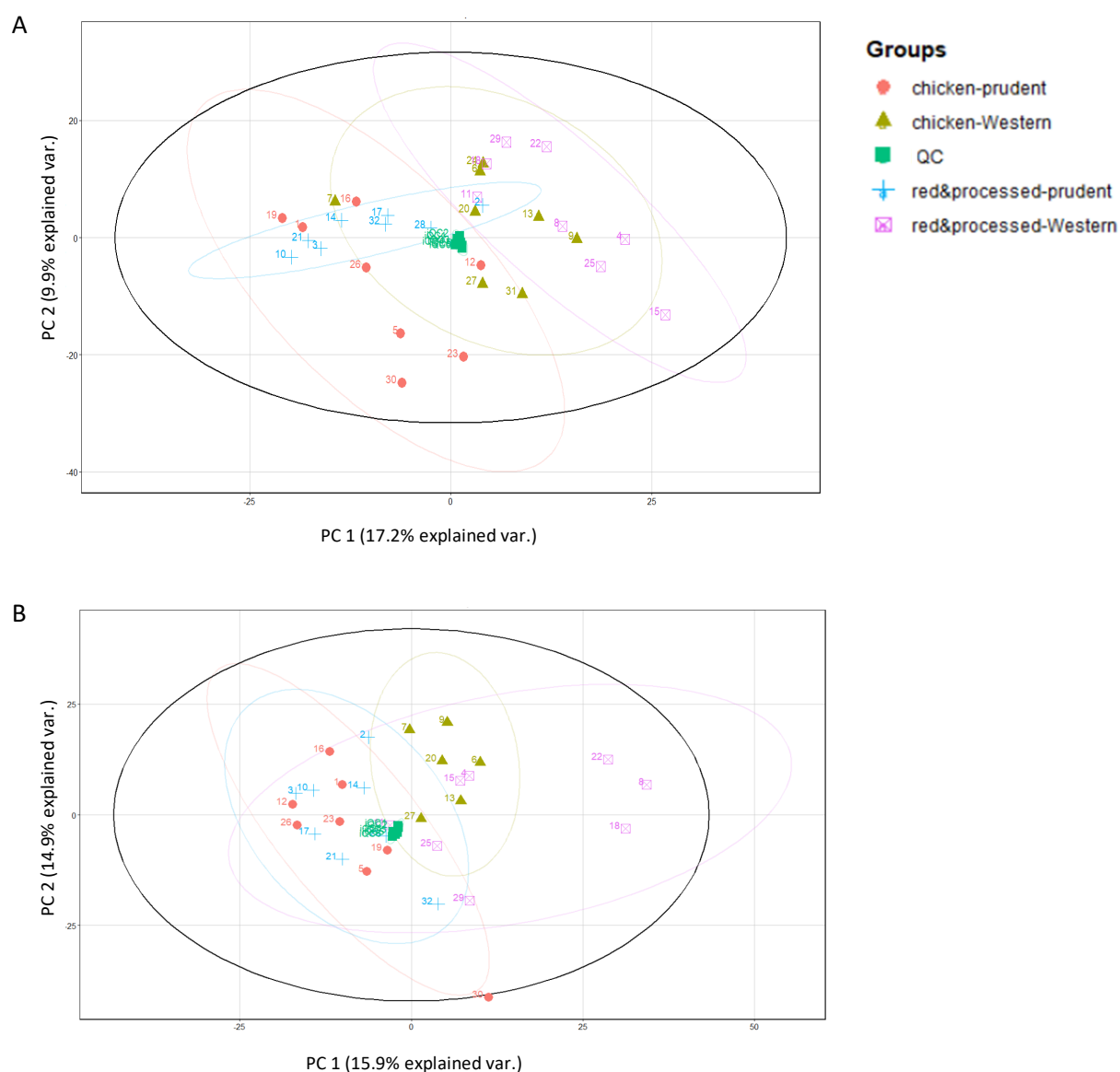


Figure S3. PCA-X score plots based on the small intestinal (A) and colon (B) metabolome of pigs fed four dietary treatments. Separation of the two background diets was observed, with the prudent diets situated at the left, and the Western diets situated at the right. The close clustering of the quality control samples (QC) indicated good instrumental performance. Data were subjected to log-transformation and Pareto scaling and only those components with a coefficient of variance lower than 30% for the QCs were included.

Table S7. Additional information on the (tentatively) identified metabolites that were influenced by meat intake in the luminal content of the small intestine (n = 8 piglets per dietary treatment).

				multivariate statistics		information for identification				
Tentative metabolite identity	m/z	RT (min)	average peak intensity	VIP ^d	q-value ^e	MS/MS fragment ions samples (relative abundancy)		Collision energy (eV)	CSI:FingerID similarity (%)	Matched fragments Metfrag (HMDB)
Chicken										
Cadaverine ^b	103.123	0.76	854496	2.9	0.016	102.128 (100) 103.131 (15) 74.097 (15) 58.066 (6) 72.081 (3)		30	ND	4/8
3- (or 1-)Methylhistidine ^a	170.092	0.85	753411	NA	NA	ND		NA	ND	ND
Acetylglutamine	173.139	1.69	198037	4.9	<0.001	173.139 (100) 114.091 (68) 156.113 (55) 113.107 (27) 115.087 (14)		30	62.72	NA
Valyl-Proline	215.139	1.77	575702	4.1	<0.001	72.081 (100) 215.139 (38) 216.142 (2) 73.085 (2)		20	77.11	3/11
Anserine ^b	241.129	0.86	1012975	5.5	<0.001	241.129 (100) 102.128 (77) 170.092 (62) 109.076 (54) 126.103 (28)		30	76.13	14/18
Aspartyl-Methionine	265.085	2.68	209304	2.8	<0.001	265.085 (100) 150.058 (91) 171.076 (77) 219.029 (74) 70.029 (37)		20	81.48	11/17
Aspartyl-Phenylalanine	281.113	5.83	657929	2.4	0.006	120.081 (100) 166.086 (53) 70.029 (41) 235.107 (34) 74.024 (11)		30	90.48	9/16
DL-alpha-Asp-Gly-DL-leu	304.150	5.55	805697	2.9	<0.001	132.102 (100) 86.097 (95) 240.134 (82) 280.867 (75) 173.056 (70)		20	76.23	NA
H-Asp-Asp-Leu-OH	362.155	5.39	699242	2.0	0.028	231.061 (100) 86.097 (41) 132.102 (39) 203.066 (33) 158.045 (32)		20	78.69	NA
H-Gly-DL-Asp-DL-Pro-DL-Leu-OH	401.202	6.64	269170	4.6	<0.001	229.154 (100) 70.066 (25) 252.098 (19) 401.202 (16) 224.102 (12)		20	70.76	NA
H-Val-Glu-Thr-Asp-Gly-OH	520.224	5.17	264789	1.9	0.024	385.134 (100) 403.145 (62) 229.118 (61) 502.214 (40) 274.103 (40)		20	52.95	NA
Litocholyltaurine	484.308	9.47	7647667	2.9	0.033	448.287 (100) 110.027 (15) 430.294 (14) 449.291 (11) 431.298 (10)		20	NA	7/20
Red&processed meat										
L-carnitine ^b	162.112	0.91	12370378	7.1	<0.001	162.112 (100) 103.039 (20) 60.082 (14) 163.116 (4) 85.029 (4)		30	83.88	5/12
C2-carnitine ^b	204.123	1.41	16799270	6.3	<0.001	85.029 (100) 204.123 (42) 60.082 (19) 145.050 (15) 144.102 (3)		30	88.49	4/12
C3-carnitine ^b	218.138	2.69	1097364	5.2	<0.001	85.029 (100) 218.138 (29) 159.065 (16) 60.082 (13) 102.055 (2)		30	85.22	5/11
C4-carnitine ^b	232.154	5.24	2570936	5.8	<0.001	85.029 (100) 232.154 (33) 173.081 (20) 60.082 (13) 70.0657 (4)		30	72.59	NA
C5-carnitine ^b	246.169	6.58	1296199	4.8	<0.001	85.029 (100) 246.169 (40) 187.096 (22) 60.082 (12) 85.065 (4)		30	84.17	4/12
C4-3-OH-carnitine	248.149	1.73	377410	5.5	<0.001	85.029 (100) 248.149 (66) 189.075 (21) 103.039 (17) 60.082 (17)		30	78.83	10/18
C6-carnitine ^b	260.185	8.22	172532	4.8	<0.001	85.029 (100) 260.185 (29) 201.112 (14) 60.082 (12) 99.081 (3)		30	84.09	5/10
C4-DC-carnitine	262.128	1.64	705597	6.9	<0.001	85.029 (100) 262.128 (74) 60.082 (26) 203.054 (14) 103.039 (8)		30	76.50	8/11
3-Dehydroxycarnitine	146.117	0.98	3460704	5.3	<0.001	84.081 (100) 87.045 (96) 130.050 (83) 84.045 (77) 146.117 (64)		20	52.01	6/16
Carnosine ^b	227.113	0.85	1523574	3.2	0.002	210.087 (100) 156.077 (97) 110.072 (90) 227.114 (60) 181.108 (37)		20	89.96	11/19
Hydroxypropyl-Leucine	245.149	1.55	365075	4.0	<0.001	245.149 (100) 70.066 (15) 246.153 (5) 86.097 (5) 100.076 (4)		30	45.85	5/17
LysoPC(O-14:1)	452.313	12.35	351349	2.9	0.002	280.299 (100) 298.310 (32) 281.303 (10) 155.010 (7) 378.277 (6)		30	60.31	NA
LysoPC(P-16:0)	480.344	11.88	2042803	2.8	0.024	104.107 (100) 86.097 (36) 184.073 (32) 480.345 (28) 181.026 (20)		30	88.32	9/18
LysoPC (P-18:0)	508.375	12.90	247191	3.4	<0.001	104.107 (100) 184.073 (17) 86.096 (8) 124.999 (5) 508.376 (4)		30	89.28	8/10
LysoPC (O-18:0)	510.391	12.96	376767	2.5	0.022	104.107 (100) 184.073 (14) 91.055 (9) 86.097 (8) 167.979 (6)		30	83.12	12/17
Red&processed meat (× Western background diet)										
C10:1-carnitine	314.232	9.39	180965	2.8	<0.001	85.029 (100) 314.232 (22) 255.159 (11) 60.082 (10) 128.143 (5)		30	70.95	13/18
C10-carnitine ^b	316.247	9.55	528536	5.0	<0.001	85.029 (100) 316.248 (27) 60.082 (12) 257.174 (10) 155.143 (4)		30	86.46	4/10
C12:1-carnitine	342.263	9.77	473147	2.8	0.012	85.029 (100) 342.263 (18) 283.190 (12) 60.081 (8) 181.159 (6)		30	66.21	15/18
C12-carnitine	344.279	9.97	821863	3.4	0.001	85.029 (100) 344.279 (23) 60.082 (12) 285.206 (7) 183.174 (3)		30	91.06	5/9
C14:2-carnitine	368.279	9.94	369617	2.6	0.014	85.029 (100) 368.279 (23) 60.082 (15) 189.163 (7) 81.070 (5)		30	73.87	14/26
C14:1-carnitine	370.294	10.22	2236296	3.9	<0.001	85.029 (100) 370.295 (22) 60.082 (15) 311.222 (5) 191.179 (4)		30	78.77	7/9
C14-carnitine ^b	372.310	10.49	962582	3.3	0.001	85.029 (100) 355.263 (30) 372.311 (25) 60.082 (13) 161.132 (7)		30	68.17	4/17
C16:1-carnitine ^c	398.325	10.69	805908	3.7	0.001	85.029 (100) 398.326 (21) 60.081 (16) 81.070 (15) 357.278 (9)		30	76.59	NA
C16-carnitine ^b	400.341	11.08	1178251	2.7	0.020	85.029 (100) 400.342 (18) 60.082 (12) 341.267 (5) 144.102 (3)		30	86.38	4/11

C18:2-carnitine	424.341	10.85	533444	2.9	0.009	85.029 (100) 424.341 (23) 60.082 (16) 69.071 (8) 83.086 (5)	30	73.77	5/13
C18:1-carnitine ^b	426.357	11.22	2646494	3.3	0.004	85.029 (100) 426.357 (16) 60.082 (12) 144.102 (3) 427.361 (2)	30	80.44	10/11
C18-carnitine ^b	428.372	11.64	560047	3.6	0.001	85.029 (100) 428.372 (21) 60.082 (12) 369.299 (5) 144.102 (3)	30	84.54	8/12
Palmitoleoylglycerol	329.268	12.85	168654	1.6	0.200	237.221 (100) 311.258 (97) 219.211 (77) 121.101 (38) 135.116 (33)	20	89.78	14/19
Palmitoylglycerol ^c	331.283	13.59	2140733	2.9	0.006	313.273 (100) 57.071 (37) 95.086 (36) 71.086 (34) 109.101 (29)	20	91.11	12/20
Linolenoylglycerol	353.268	12.55	280085	2.3	0.023	184.073 (100) 103.076 (48) 81.019 (28) 194.115 (21) 165.113 (15)	20	49.01	6/20
Oleoylglycerol ^b	357.299	13.80	1203880	1.3	0.701	81.070 (100) 95.086 (96) 69.071 (95) 83.086 (66) 67.055 (65)	40	94.24	13/18
Stearoylglycerol ^c	359.315	14.67	211623	1.4	0.231	341.304 (100) 57.071 (35) 109.102 (34) 95.086 (32) 267.268 (31)	20	91.49	11/19

a) Metabolites were not selected based on untargeted statistical analysis, but were added based on literature or obtained results in the other gastrointestinal compartment. b) Identification was confirmed with commercial standards. c) The identity of the detected metabolite does not correspond with the available specific standard, but is likely an isomer of the available standard, which is covered by the nomenclature used in the list d) VIP scores obtained from OPLS-DA models comparing chicken versus red and processed meat. e) Benjamini Hochberg adjusted p-values (q-value) to correct for false discovery rates obtained in *limma* investigating the main effect meat (chicken vs. red and processed meat). NA= not available, ND=not determined. *m/z* corresponds with the accurate [M+H]⁺ mass of the parent ion.

Table S8. Additional information on the (tentatively) identified metabolites that were influenced by meat intake in the luminal content of the colon (n = 8 piglets per dietary treatment, except chicken-western, where n = 6).

				multivariate statistics		information for identification							
tentative metabolite identity	m/z	RT (min)	average peak intensity	VIP ^d	q-value ^e	MS/MS fragment ions samples (relative abundancy)					Collision energy (eV)	CSI:FingerID similarity (%)	Matched fragments Metfrag (HMDB)
Chicken													
2-(4-Methylimidazol-1-yl)ethanol	127.087	1.01	1595392	5.4	<0.001	127.087 (100)	109.076 (11)	96.069 (6)	128.090 (5)	126.066 (3)	30	47.34	NA
4-Imidazolone-5-propionic acid	157.061	2.45	301947	2.0	0.016	157.061 (100)	106.992 (66)	125.002 (24)	78.998 (18)	111.056 (17)	30	40.28	4/17
Guanidinovaleric acid	160.108	1.88	946166	4.5	<0.001	160.108 (100)	101.060 (27)	100.076 (15)	118.086 (5)	128.951 (5)	30	72.53	NA
Methylglutamate	162.076	1.89	1743357	5.3	<0.001	144.065 (100)	102.055 (24)	72.045 (13)	126.055 (13)	90.055 (12)	30	52.85	11/17
3- (or 1-)Methylhistidine	170.092	0.89	447080	2.0	0.014	170.092 (100)	152.071 (60)	109.076 (30)	126.103 (21)	96.069 (18)	20	91.9	7/17
N-(4,5-Dihydro-1-methyl-4-oxo-1H-imidazol-2-yl)alanine	186.087	1.04	2269198	2.1	0.028	186.087 (100)	140.082 (12)	163.940 (7)	117.980 (7)	128.951 (5)	30	50.65	5/17
Prolylserine	203.103	2.30	570015	3.9	<0.001	114.055 (100)	90.055 (16)	126.055 (15)	185.092 (14)	203.102 (10)	20	45.31	8/17
Acetyl-3-(or 1-)methylhistidine	212.103	1.03	505607	3.1	<0.001	212.103 (100)	109.076 (61)	91.039 (33)	85.029 (29)	145.049 (22)	30	62.76	
2-(3-Carboxy-3-aminopropyl)-L-histidine	257.124	1.48	380016	3.1	<0.001	168.077 (100)	257.124 (62)	258.094 (26)	114.066 (24)	126.055 (10)	20	35.4	6/18
H-Pro-Val-Val-OH	314.207	5.96	436362	3.8	<0.001	314.207 (100)	268.201 (35)	110.072 (24)	156.077 (22)	315.212 (8)	30	52.52	NA
deamino-hPhe-Ala-Ala-Pro-NH2	403.234	9.13	325146	4.6	<0.001	293.186 (100)	168.102 (39)	403.233 (31)	86.097 (20)	180.109 (18)	20	49.81	NA
Suberic acid	175.097	8.05	1119094	2.0	0.566	83.086 (100)	157.086 (43)	111.081 (39)	175.097 (22)	55.055 (18)	20	83.94	9/19
C ₁₁ H ₂₀ O ₄ (Undecanedioic acid) ^b	217.143	9.53	134362	2.9	0.003	135.117 (100)	153.127 (62)	86.097 (40)	69.071 (38)	217.143 (27)	20	73.73	7/17
C ₁₂ H ₁₈ O ₄ (Dioxo-dodecenoic acid)	227.128	9.14	331716	2.5	0.003	111.044 (100)	181.122 (71)	163.112 (62)	153.127 (48)	135.117 (31)	30	53.82	NA
C ₁₂ H ₂₂ O ₅ (Hydroxydodecanedioic acid)	247.154	8.99	284416	3.1	<0.001	229.143 (100)	211.132 (25)	147.117 (19)	193.122 (16)	165.127 (15)	20	65.16	NA
C ₁₈ H ₃₀ O ₂ (Octadecatrienoic acid)	279.232	10.16	292477	3.3	<0.001	117.070 (100)	149.023 (66)	279.231 (46)	279.158 (30)	95.086 (21)	20	62.22	8/14
C ₁₈ H ₃₀ O ₆ ((-)-11-Hydroxy-9,15,16-trioxooctadecanoic acid)	343.211	9.69	258061	2.5	<0.001	343.199 (100)	133.101 (35)	125.096 (33)	97.065 (28)	109.101 (26)	30	50.63	9/18
Glucosyl (2E,6E,10X)-10,11-dihydroxy-2,6-farnesadienoate	433.245	9.37	293184	2.8	<0.001	293.211 (100)	414.230 (80)	146.027 (65)	432.241 (53)	415.233 (48)	20	46.76	5/18
Chicken (× Western background diet)													
H-DL-Leu-Gly-DL-Glu-OH	318.166	4.89	346464	1.1	0.315	319.136 (100)	143.118 (49)	148.060 (18)	86.097 (17)	130.050 (5)	20	72.54	NA

Cyclo(L-Valyl-L-Leucyl)	213.160	7.94	548245	1.4	0.544	213.160 (100) 72.081 (76) 86.097 (74) 140.143 (30) 168.138 (29)	30	61	NA
Cyclo(L-Valyl-L-Phenylalanyl)	247.144	8.69	134999	0.5	0.568	120.081 (100) 72.081 (45) 247.144 (29) 174.128 (18) 219.149 (18)	30	57.78	NA
Cyclo(L-Leucyl-L-Phenylalanyl)	261.160	9.24	845659	0.2	0.805	120.081 (100) 86.097 (43) 261.159 (25) 216.138 (16) 233.164 (15)	30	62.19	5/18
Chicken (× prudent background diet)									
C ₁₈ H ₃₂ O ₃ (HODE) ^c	295.228	10.90	1493774	1.6	0.456	295.228 (100) 296.231 (11) 230.986 (3) 134.894 (3) 158.977 (2)	30	79.38	15/18
C ₁₈ H ₃₄ O ₃ (Hydroxyoctadecenoic acid)	299.258	10.60	719699	2.2	0.234	263.237 (100) 245.226 (91) 97.101 (87) 83.086 (62) 111.117 (58)	20	79.64	13/20
C ₁₈ H ₃₆ O ₄ (Dihydroxyoctadecanoic acid)	317.268	10.61	1001222	2.2	0.302	263.237 (100) 97.102 (76) 245.226 (72) 83.086 (52) 111.117 (47)	20	75.29	6/20
Red&processed meat									
3-Dehydroxycarnitine ^a	146.117	0.96	55768092	1.5	0.349	146.117 (100) 87.044 (52) 60.082 (34) 147.121 (7) 84.081 (4)	20	73.69	5/17
L-carnitine ^a	162.112	0.93	4751179	1.3	0.437	ND	NA	ND	ND
C3-carnitine	218.139	3.34	582040	2.4	0.036	85.029 (100) 218.138 (29) 159.065 (16) 60.082 (13) 102.055 (2)	30	85.22	5/11
C4-DC-carnitine	262.128	1.59	827900	1.3	0.024	85.029 (100) 262.128 (74) 60.082 (26) 203.054 (14) 103.039 (8)	30	76.50	8/11
C ₁₈ H ₃₂ O ₄ (HPODE) ^c	311.223	10.96	714170	2.6	0.021	311.223 (100) 311.168 (79) 171.102 (52) 293.212 (48) 211.133 (32)	30	84.62	13/16
C ₁₈ H ₃₄ O ₅ (TriHOME)	329.234	9.65	1843683	1.9	0.039	329.233 (100) 171.101 (26) 330.236 (14) 229.144 (13) 211.133 (13)	30		8/15
C ₁₈ H ₃₀ O ₃ (15(16)-epODE) ^a	295.226	9.66	622158	1.2	0.523	ND	NA	ND	ND
D-pipecolic acid ^b	130.086	1.04	16366507	2.7	0.032	84.081 (100) 130.086 (37) 84.045 (34) 70.066 (4) 130.035 (4)	40	63.41	6/11
2-amino-6-Hydroxyhexanoic acid	148.097	0.98	1470157	2.5	0.006	84.045 (100) 130.050 (42) 102.055 (38) 148.061 (12) 147.121 (9)	20	65.66	NA
C ₁₄ H ₂₆ O ₃ (Oxotetradecanoic acid)	243.195	11.87	284812	2.9	0.003	95.086 (100) 109.101 (78) 83.086 (36) 165.164 (35) 107.967 (31)	30	63.19	7/19
Norecasantalic acid or Jasmolone	181.122	10.13	381323	2.1	0.001	181.122 (100) 163.112 (11) 135.117 (11) 182.126 (6) 116.972 (4)	30	50.55	13/18
Hydroxypropyl-Leucine	245.149	1.55	1229321	5.9	<0.001	245.149 (100) 246.153 (7) 60.082 (5) 100.076 (3) 70.066 (3)	30	41.47	8/18
Hydroxypropyl-Asparagine	246.108	1.42	5567373	2.8	0.001	246.108 (100) 132.077 (22) 186.087 (19) 228.098 (7) 246.1319 (6)	30	34.31	7/16

a) Metabolites were not selected based on untargeted statistical analysis, but were added based on literature or obtained results in the other gastrointestinal compartment. b) Identification was confirmed with commercial standards. c) The identity of the detected metabolite does not correspond with the available specific standard, but is likely an isomer of the available standard, which is covered by the nomenclature used in the list d) VIP scores obtained from OPLS-DA models comparing chicken versus red and processed meat. e) Benjamini Hochberg adjusted p-values (q-value) to correct for false discovery rates obtained in *limma* investigating the main effect meat (chicken vs. red and processed meat). NA= not available, ND=not determined. *m/z* corresponds with the accurate [M+H]⁺ mass of the parent ion, except for C₁₈H₃₂O₃ (HODE), C₁₈H₃₂O₄ (HPODE) and C₁₈H₃₄O₅ (TriHOME), where *m/z* corresponds with [M-H]⁻.

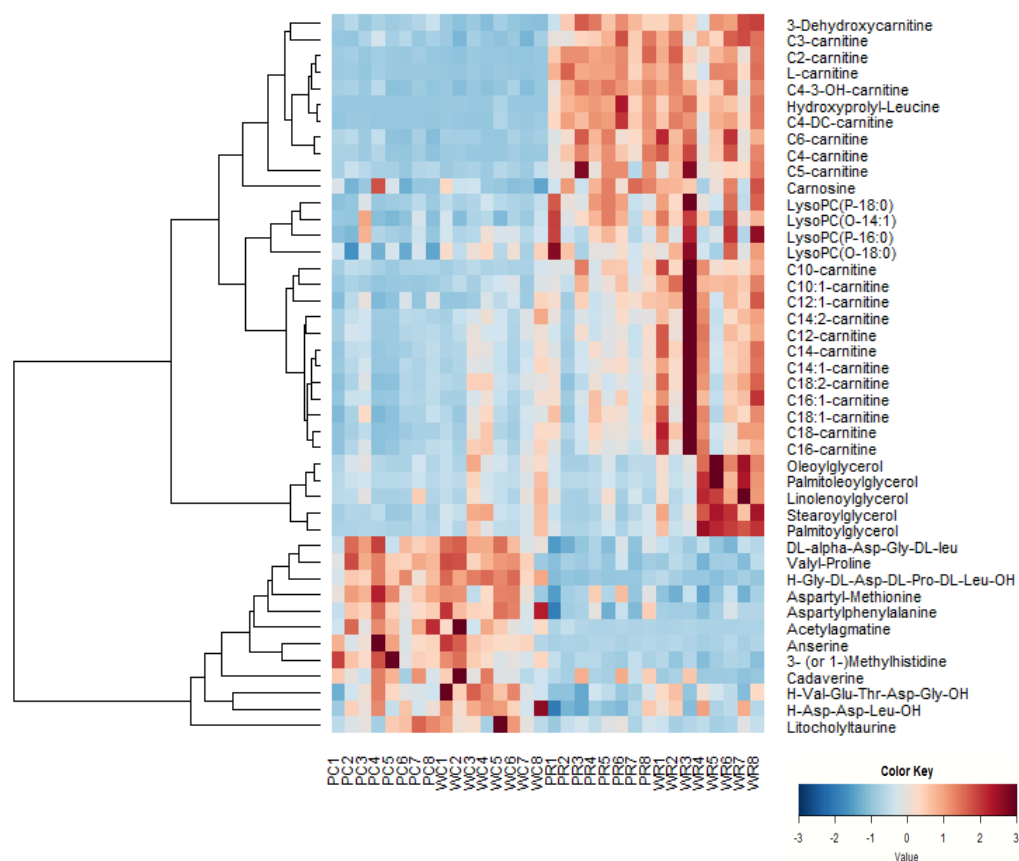


Figure S4. Heatmap of normalized peak intensities of (tentatively) characterized meat-associated metabolites in the luminal content of the small intestine. Columns represent samples (n=8 per dietary treatment) and rows represent metabolites. Data was by default scaled to limits of -3,3. PC=chicken-prudent, WC=chicken-Western, PR=red&processed-prudent, WR=red&processed-Western.

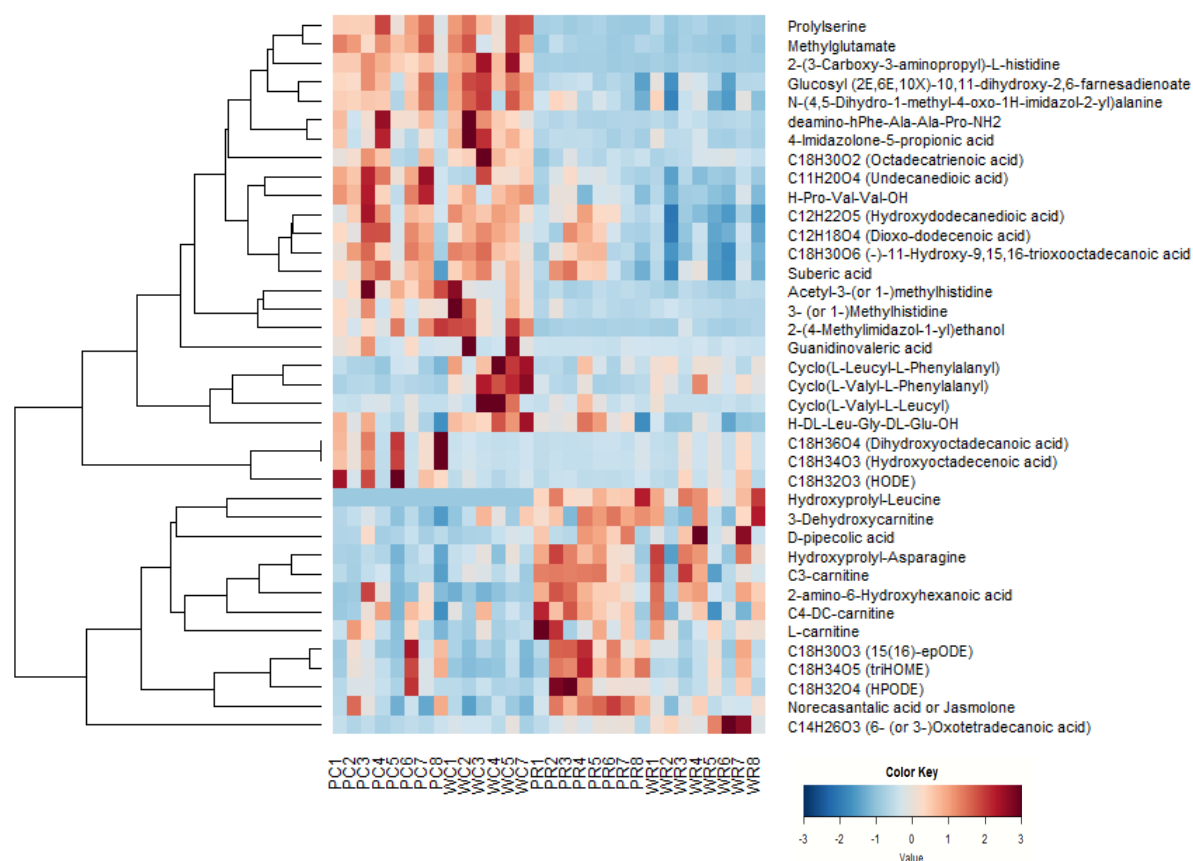


Figure S5. Heatmap of normalized peak intensities of (tentatively) characterized meat-associated metabolites in the luminal content of the colon. Columns represent samples (n = 8 per dietary treatment, except chicken-western, where n = 6) and rows represent metabolites. Data was by default scaled to limits of -3,3. PC=chicken-prudent, WC=chicken-Western, PR=red&processed-prudent, WR=red&processed-Western.

Table S9. Fatty acid composition of the diets expressed as mg fatty acid/100 g diet

	prudent		Western	
	chicken	red&processed	chicken	red&processed
C10:0	28	28	49	48
C12:0	38	33	96	84
C14:0	94	102	202	198
C15:0	7	11	20	16
C16:0	643	711	1369	1433
C17:0	8	9	17	19
C18:0	180	268	434	545
C20:0	7	9	16	17
C22:0	3	4	14	13
C24:0	1	0	1	1
Total SFA	1010	1175	2218	2375
C14:1	8	7	19	18
C16:1	74	52	108	85
C17:1	4	5	6	9
c9C18:1	1246	1238	2054	2155
c11C18:1	62	74	96	116
C20:1	9	13	16	25
C22:1	1	1	1	2
C24:1	2	2	2	2
Total MUFA	1406	1392	2303	2412
C18:2n-6	557	507	1134	1077
C18:3n-6	2	1	3	2
C20:2n-6	4	8	5	11
C20:3n-6	3	4	5	5
C20:4n-6	11	11	15	17
C22:4n-6	3	4	3	4
C22:5n-6	2	2	2	2
Total n-6 PUFA	581	536	1167	1117
C18:3n-3	87	96	110	122
C20:3n-3	2	4	3	5
C20:4n-3	1	1	1	1
C20:5n-3	1	2	2	4
C22:5n-3	3	4	4	7
C22:6n-3	1	1	1	1
Total n-3 PUFA	96	109	121	141
Total	3092	3212	5810	6045